Answer 1:

Bibliographic Information

Development of novel 68Ga- and 18F-labeled GnRH-I analogs with high GnRHR-targeting efficiency. Schottelius, Margret; Berger, Sebastian; Poethko, Thorsten; Schwaiger, Markus; Wester, Hans-Juergen. Nuklearmedizinische Klinik and Poliklinik, Klinikum rechts der Isar, Technische Universitaet Muenchen, Munich, Germany. Bioconjugate Chemistry (2008), 19(6), 1256-1268. Publisher: American Chemical Society, CODEN: BCCHES ISSN: 1043-1802. Journal written in English. CAN 149:153355 AN 2008:647293 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

Abstract

A large majority of tumors of the reproductive system express the gonadotropin releasing hormone receptor (GnRHR). Blockade and activation of this receptor with various antagonistic and agonistic analogs of native GnRH-I (pGlu1-His2-Trp3-Ser4-Tyr5-Gly6-Leu7-Arg8-Pro9-Gly10-NH2), resp., has shown efficient suppression of tumor growth. In this study, the GnRH-receptor system has been evaluated with respect to its suitability as a target for in vivo peptide receptor targeting using radiolabeled GnRH-analogs, and in parallel, new 18F- and 68Ga-labeled GnRH analogs have been developed. In vitro radioligand binding assays performed with various GnRHR-expressing human cell lines using [125I]Triptorelin (D-Trp6-GnRH-I) as the std. radioligand revealed a very low level of GnRH receptor expression on the cell surface. Generally, total cellular activity was very low (.apprx.3% of the applied activity), and only a small fraction (max. 40%) of cell-assocd. activity could be attributed to receptor-specific radioligand binding/internalization. However, substitution of fetal calf serum by NU serum in the culture medium led to increased and stable GnRHR-expression, esp. in the ovarian cancer cell line EFO-27, thus allowing for a stable exptl. setup for the evaluation of the new radiolabeled GnRH-I analogs. The new radiolabeled GnRH-I analogs developed in this study were all based on the D-Lys6-GnRH-I-scaffold. For 68Ga-labeling, the latter was coupled with DOTA at D-Lys6. To allow 18F-labeling via chemoselective oxime formation, D-Lys6-GnRH-I was also conjugated with Ahx (aminohexanoic acid) or β-Ala, which in turn was coupled with Boc-aminooxyacetic acid. 18F-labeling via oxime formation with 4-[18F]fluorobenzaldehyde was performed using the Boc-protected (Boc = tert-butoxycarbonyl) precursors. Receptor affinities of [68Ga]DOTA-GnRH-I, D-Lys6-Ahx([18F]FBOA)-GnRH-I, and D-Lys6-βAla([18F]FBOA)-GnRH-I (FBOA = fluorobenzyloxime acetyl) were detd.

using GnRHR-membrane prepns., and internalization efficiency of the new radioligands was detd. in EFO-27 cells. Both quantities were highest for D-Lys6-Ahx([18F]FBOA)-GnRH-I (IC50 = 0.50 ± 0.08 nM vs 0.13 ± 0.08 nM for Triptorelin; internalization: 86 ± 16% of the internal ref. [125I]Triptorelin), already substantially reduced in the case of the -βAla([18F]FBOA)-deriv. (IC50 = 0.86 ± 0.13 nM; internalization: 42 ± 3% of [125I]Triptorelin), while the [68Ga]DOTA-analog showed almost complete loss of binding affinity and ligand internalization (IC50 = 13.3 ± 1.0 nM; internalization: 2.6 ± 1.0% of [125I]Triptorelin). Generally, the lipophilic residue [18F]FBOA is much better tolerated as a modification of the D-Lys6-side chain, with receptor affinity of the resp. analogs strongly depending upon spacer length between the D-Lys6-side chain and the [18F]FBOA-moiety. In summary, D-Lys6(Ahx-[18F]FBOA)-GnRH-I shows the highest potential for efficient GnRHR-targeting in vivo of the compds. investigated. Unfortunately, however, the very low cell surface expression of GnRH-receptors and thus very low radioligand uptake by GnRHR-pos. tumor cells found in vitro was also confirmed by a preliminary biodistribution study in OVCAR-3 xenografted nude mice using the std. GnRHR radioligand [125I]Triptorelin. Tumor uptake was lower than blood activity concn. at 1 h p.i. (0.49 ± 0.05 vs 0.96 ± 0.13 for tumor and blood, resp.). These data seriously challenge the suitability of the GnRHR-system as a suitable target for in vivo peptide receptor imaging using radiolabeled GnRH-I derivs., despite the availability of high-affinity radiolabeled receptor-ligands such as D-Lys6(Ahx-[18F]FBOA)-GnRH-I.

Answer 2:

Bibliographic Information

Down-regulation and change in subcellular distribution of receptors for luteinizing hormone-releasing hormone in OV-1063 human epithelial ovarian cancers during therapy with LH-RH antagonist Cetrorelix. Halmos, Gabor; Schally, Andrew V.; Kahan, Zsuzsanna. Endocrine, Polypeptide and Cancer Institute, Veterans Affairs Center and Department of Medicine, Tulane University School of Medicine, New Orleans, LA, USA. International Journal of Oncology (2000), 17(2), 367-373. Publisher: International Journal of Oncology, CODEN: IJONES ISSN: 1019-6439. Journal written in English. CAN 133:261736 AN 2000:566696 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

Abstract

The inhibition of growth of various hormone-dependent cancers by analogs of LH-releasing hormone (LH-RH) may be exerted in part through receptors for LH-RH present on tumor cells, but the direct mode of action of LH-RH agonists and antagonists is still not completely understood. The aim of this study was to investigate the effects of agonist [D-Trp6]LH-RH and antagonist Cetrorelix, administered s.c. at a dose of 100 μg/day for 3 wk on the binding characteristics and subcellular localization of receptors for LH-RH in OV-1063 human epithelial ovarian cancers xenografted into nude mice. Using radioligand binding studies, following in vitro desatn., we demonstrated the presence of specific, high affinity binding sites for LH-RH in both cell membrane and nuclear fraction of OV-1063 tumors. Treatment with Cetrorelix, but not [D-Trp6]LH-RH, caused about 60% redn. ($p<0.01$) in tumor vol. and wt. [D-Trp6]LH-RH decreased the no. of LH-RH receptors on OV-1063 tumor membranes by 44% after 14 days ($p<0.01$), and the concn. of receptors remained at that level on day 21. The maximal binding capacity of receptors for LH-RH in the nuclei was significantly higher ($p<0.05$) after 3 wk of treatment with [D-Trp6]LH-RH. Cetrorelix decreased the concn. of membrane receptors for LH-RH by 53% ($p<0.01$) after 14 days and the levels on day 21 were even lower, showing a 70% redn. ($p<0.01$). In contrast, the no. of LH-RH binding sites in the nuclear pellet was significantly increased ($p<0.01$) by Cetrorelix at that time. Our results demonstrate for the first time that the down-regulation of LH-RH receptors on the cell membranes of OV-1063 human ovarian cancers after therapy with antagonist Cetrorelix or agonist [D-Trp6]LH-RH is assocd. with an increase in receptor concn. in the nuclei. These phenomena could be related to the internalization and subcellular translocation of receptors in these tumor cells.

Answer 3:

Bibliographic Information

Decrease in the level and mRNA expression of LH-RH and EGF receptors after treatment with LH-RH antagonist Cetrorelix in DU-145 prostate tumor xenografts in nude mice. Lamharzi, Najib; Halmos, Gabor; Jungwirth, Andreas; Schally, Andrew V. Endocrine, Polypeptide and Cancer Institute, Veterans Affairs Medical Center and Department of Experimental Medicine, Tulane University School of Medicine, New Orleans, LA, USA. International Journal of Oncology (1998), 13(3), 429-435. Publisher: International Journal of Oncology, CODEN: IJONES ISSN: 1019-6439. Journal written in English. CAN 129:311197 AN 1998:566559 CAPLUS

Abstract

Using radioligand binding, RT-PCR, and Southern blot analyses, the authors evaluated whether agonist [D-Trp6]LH-RH and antagonist Cetrorelix could affect the levels of receptors for LH-RH and EGF and expression of mRNA for these receptors in DU-145 human androgen-independent prostate cancers xenografted into nude mice. Radioligand binding studies showed the presence of specific high affinity receptors for LH-RH and EGF in DU-145 prostate tumors. Cetrorelix, but not [D-Trp6]LH-RH significantly inhibited tumor growth. The concn. of LH-RH receptors was reduced by 22% and 67% after 4 wk of treatment with [D-Trp6]LH-RH and Cetrorelix resp. The concn. of EGF receptors fell by 48% in the [D-Trp6]LH-RH group, whereas Cetrorelix led to a 66% redn. The expression of LH-RH and EGF receptor mRNA was investigated by RT-PCR anal. followed by Southern blotting. Densitometric anal. of the developed bands showed that the antagonist Cetrorelix decreased the expression of LH-RH receptor mRNA by 55% compared to control group while the 20% redn. after treatment with the LH-RH agonist was non-significant. Treatment with [D-Trp6]LH-RH and Cetrorelix also reduced the expression of EGF receptor mRNA by 35% and 68% resp. compared to control group. In conclusion, these data demonstrate that growth inhibition of DU-145 prostate tumors induced by prolonged administration of LH-RH antagonist Cetrorelix is accompanied by a marked decrease in the concn. of LH-RH and EGF receptors as well as in their mRNA levels.

Answer 4:

Bibliographic Information

Inhibition of growth of androgen-independent DU-145 prostate cancer in vivo by luteinizing hormone-releasing hormone antagonist cetrorelix and bombesin antagonists RC-3940-II and RC-3950-II. Jungwirth, A.; Pinski, J.; Galvan, G.; Halmos, G.; Szepeshazi, K.; Cai, R. Z.; Groot, K.; Vadillo-Buenfil, M.; Schally, A. V. Department of Experimental Medicine, Veterans Affairs Medical Center, Tulane University School of Medicine, New Orleans, LA, USA. European Journal of Cancer (1997), 33(7), 1141-1148. Publisher: Elsevier, CODEN: EJCAEL ISSN: 0959-8049. Journal written in English. CAN 127:144862 AN

1997:477134 CAPLUS (Copyright (C) 2008 ACS on SciFinder (R))

Abstract

The aim of this study was to test the antagonist of LH-RH (Cetrorelix), agonist [D-Trp6]LH-RH (triptorelin) and new bombesin antagonists RC-3940-II and RC-3950-II for their effect on the growth of an androgen-independent prostate cancer cell line, DU-145, xenografted into nude mice. Xenografts were grown in male nude mice, and after 4 wk, the animals were treated either with saline (control) or with one of the analogs. One group of mice was given a combination of Cetrorelix and RC-3950-II. Treatment was given for 4 wk. Tumor and body wts., and tumor vols. were measured. At sacrifice, tumors were dissected for histol. examn. and receptor studies. Serum was collected for measurement of hormone levels. The final tumor vol. in control animals injected with saline was 577 ± 155 mm3 and that of animals treated with Cetrorelix only 121.4 ± 45 mm3 ($P < 0.01$). Bombesin antagonists RC-3940-II and RC-3950-II also significantly reduced DU-145 tumor vol. in nude mice to 84.9 ± 19.9 and 96.8 ± 28 mm3, resp. Agonist [D-Trp6]LH-RH did not significantly inhibit tumor growth. Serum levels of LH were decreased to 0.08 ± 0.02 ng/mL ($P < 0.05$) in the Cetrorelix treated group as compared to 1.02 ± 0.1 ng/mL for the controls, and testosterone levels were reduced to castration levels (0.01 ± 0.01 ng/mL). Specific receptors for EGF and LH-RH in DU-145 tumors were significantly downregulated after treatment with Cetrorelix, RC-3940-II and RC-3950-II. Although LH-RH could be a local regulator of growth of prostate cancer, the fall in LH-RH receptors is not fully understood and the inhibitory effects of Cetrorelix and bombesin antagonists on DU-145 tumor growth might be attributed at least in part to a downregulation of EHF receptors. Since Cetrorelix and bombesin antagonists inhibit growth of androgen-independent DU-145 prostate cancers, these compds. could be considered for the therapy of advanced prostate cancer in men, esp. after relapse.